Complete Genome Sequence of a Double-Stranded RNA Virus from Avocado

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A number of avocado (Persea americana) cultivars are known to contain high-molecular-weight double-stranded RNA (dsRNA) molecules for which a viral nature has been suggested, although sequence data are not available. Here we report the cloning and complete sequencing of a 13.5-kbp dsRNA virus isolated from avocado and show that it corresponds to the genome of a new species of the genus Endornavirus (family Endornaviridae), tentatively named Persea americana endornavirus (PaEV).

Endornaviruses (genus Endornavirus) are double-stranded RNA (dsRNA) viruses that infect plants, fungi, and oomycetes which have recently been given family status (Endornaviridae) (1). The species that infect plants do not form vires, are present in every tissue, lack cell-to-cell movement, and are transmissible only vertically at a very high rate (2). The genomes of endornaviruses encode a single long open reading frame (ORF), and most of them have a discontinuity (nick) near the 5' end of the plus strand (2). The putative polyprotein encoded by the single ORF contains a conserved RNA-dependent RNA polymerase domain (RdRp) and other domains that are not conserved among the members (12). There are 11 complete endornavirus sequences corresponding to nine recognized or putative species available: four that infect plants (5, 6, 7, 10), four that infect fungi (8, 9, 13, 14), and one that infects an oomycete (3).

A number of avocado (Persea americana) cultivars were reported to contain high-molecular-weight dsRNA molecules for which a viral nature has been suggested (4). Here we report the complete sequence of a 13.5-kbp dsRNA virus isolated from the avocado cultivar Fuerte and provide evidence that supports placing it in the genus Endornavirus. We propose the name Persea americana endornavirus (PaEV).

dsRNA was purified from total nucleic acid extracts by column chromatography as previously described (15) from mature leaves of an avocado Fuerte tree maintained at the ex situ germplasm collection of the Estación Experimental “La Mayora” (Málaga, Spain). Twenty-six overlapping cDNA clones that covered the entire molecule were obtained by a combination of different methods, including the use of random hexanucleotides, specific primers, polyadenylation and 5' rapid amplification of cDNA ends (RACE). Both strands of all clones were sequenced using an ABI 3730xl DNA analyzer, obtaining 2 times to 8 times coverage for each strand. The PaEV genome is 13,459 nucleotides (nt) in length, with 5' and 3' untranslated regions (UTRs) of 296 nt and 45 nt [including a poly(C) tail of 14 residues], respectively. A single ORF was predicted to encode a polyprotein of 501 kDa. In addition to the conserved RdRp domain, this protein contains two glycosyltransferase domains (one belonging to a superfamily that contains a DXD motif and another a GTB-type) and a helicase domain. Overall, the PaEV genome showed the highest nucleotide identity (ca. 50%) with the genomes of Oryza sativa endornavirus (GenBank accession no. D32136) and Oryza rufipogon endornavirus (GenBank accession no. AB014344). The PaEV sequence increases the known complexity and confirms the dynamic evolutionary history of the family Endornaviridae, whose plant-infecting members maintain an atypically persistent lifestyle (11, 12).

Nucleotide sequence accession number. The complete genome sequence of PaEV isolate Fuerte was deposited in GenBank under accession no. JN880414.

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